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# NOTES ON A MICROSPORIDIAN PARASITE OF A NEMATODE *

R. KUDO AND D. C. HETHERINGTON

Little is known concerning the parasites of the nematode. Up to the present five authors have noted what they regarded as parasites in *Ascaris mystax* of the cat. In the course of study on this nematode, Bischoff (1855) found in the reproductive organ peculiar bodies which he was inclined to regard as spermatozoa. These bodies were yellowish, sharply-contoured and highly refractive, measuring 7.7μ by 4.4μ. Munk (1858) noticed similar bodies in the same organ of the same host. Although these bodies varied from 4.2 to 5.9μ in length and 1.7 to 2.1μ in breadth, the author held them to be identical with those observed by Bischoff and considered them as parasitic algae, stating that "die ovalen Körperchen, wo sie in den Geschlechtskanälen der Männchen vorhanden sind, die Entwicklung der Samenkörperchen und dadurch mittelbar die Befruchtung und weitere Entwicklung der Eier verhindern."

Keferstein (1862) recognized apparently a similar parasite in the intestine and the reproductive organs of this nematode and believed it to be a fungus, naming it *Mucor helminthophthorus.* The oval spores measured 4 to 5μ long by about 2μ broad. Frequently they were surrounded by a membrane and formed a spherical body of about 20μ in diameter. He found a fungus typical to the genus Mucor in the host intestine, which varied from 4 to 20μ in diameter, and considered that the spores enclosed in a membrane were produced by the fungus. Judging from his drawings which were magnified 590 diameters and from the fact that he did not see the spherical bodies containing spores attached to the fungus, it is open to question if the stages described by him belonged to a single form of microorganism.

Lutz and Splendore (1908) described a microsporidian under the name of *Nosema mystacis* which was found parasitic in the intestine and the reproductive organs of *Ascaris mystax* of Brazilian cats. The spores were regularly oval in shape, refractive and showed a round vacuole at the posterior end, measuring 4 to 4.5μ long by 2 to 2.5μ broad. The true nature of the parasites quoted here is unknown. The microsporidian designation of the parasite by the last named two authors is open to discussion since their description of the form is very incomplete.

While examining *Protospirura muris,* a parasitic nematode of the common house mouse, *Mus musculus,* our attention was called to an apparent protozoan parasite in the epithelial cells of the intestine. A study of the spores in fresh as well as stained condition and under pressure revealed the fact that the protozoan was a microsporidian for which the name *Thelohania reniformis* nov. spec. is proposed. In view of the circumstance that this is a case of true microsporidian infection in a nematode, it seems worth while to state our observations upon it briefly.

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The host nematodes were found in the stomachs of the common house mice caught in the stables and the dairy barns of the University of Illinois at Urbana. The protozoan was found in the epithelial cells of the intestine of the host worm throughout its entire length save the very extremities and isolated spores as well as those still enclosed in the sporont membrane were also noticed in the lumen of the organ. No other organs of the host nematode showed any degree of infection by the parasite, although the body and particularly the reproductive organs were searched for evidence.

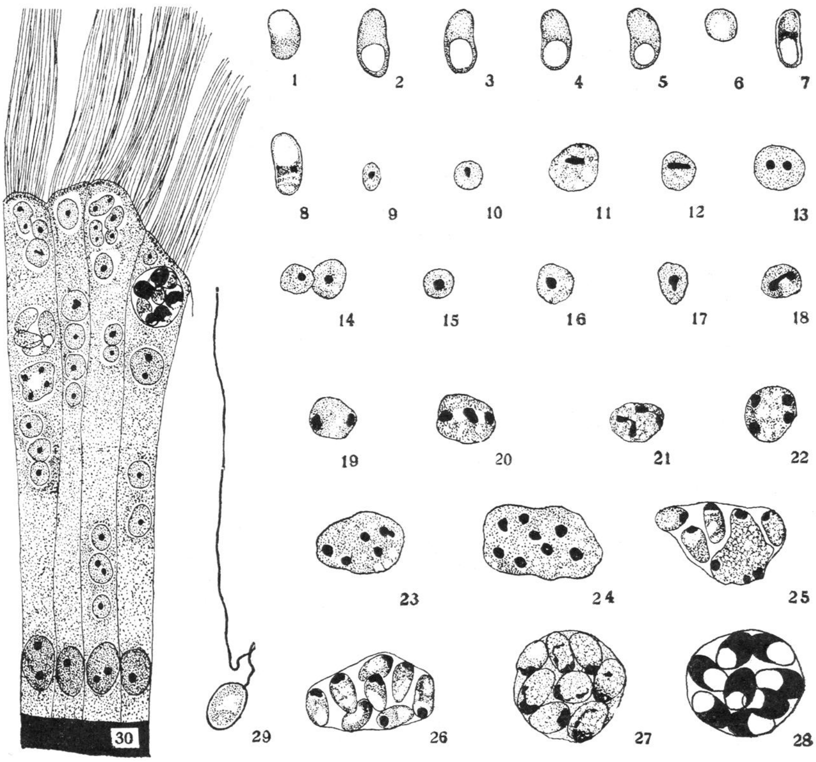
The protozoan was studied in fresh as well as stained smears and in sections. For fixation, sublimate-alcohol-acetic acid mixture, Carnoy's or Fleming's fluid, was used, while Dobell's alcoholic iron hematein, Heidenhain's iron hematoxylin or Giemsa's solution was used for staining. For the demonstraton of the polar filament of the spore, Kudo's method (Kudo, 1920) was employed.

### THE SPORE

The spores are kidney-bean shaped (Figs. 1-5) in front view; circular in cross-section (Fig. 6). The spore membrane is comparatively thin and therefore the spores are not so refractive as those of *Nosema bombycis, N. apis, Thelohania opacita* and others. Under an immersion objective the contents of the spore are found to be differentiated into two regions: near one of the extremities, which is frequently more rounded than the other, there is to be seen a rounded clear area, while the remaining portion is finely granulated. The spores measure 3 to 4μ (3.4μ average) long by 1.5 to 1.8μ broad. When a fresh spore is subjected to the action of mechanical pressure (Kudo, 1920: 87), the polar filament becomes extruded. It is comparatively short and thick, measuring 45 to 55μ long in an apparently fully extruded state (Fig. 29).

When the spore is fixed and stained, the clear space mentioned above remains unstained. Along the inner margin of this space, the cytoplasm seems to be accumulated in a zone in which one or two deeply stained nuclear grains are located. The space between this and the other extremity of the spore is occupied by a less deeply stained cytoplasmic mass (Figs. 7, 8). The structure of the spore mentioned here differs from that of *Thelohania magna* (Kudo, 1921), but is somewhat similar to that of *Nosema bombycis* or *Nosema baetis* (Kudo, 1921).

Early stages of the schizogony could only be studied in carefully made section preparations. The stages described here were observed exclusively in the epithelial cells of the host intestine such as shown in figure 30. The youngest schizont is a small oval or rounded mass of finely granulated cytoplasm containing a single nucleus (Fig. 9) and is embedded in the cytoplasm of the host cell. As it grows in size, there becomes visible a narrow clear space between it and the surrounding cytoplasm (Fig. 30). The nucleus is a deeply staining chromatin granule as is ordinarily observed in the schizont of many other Microsporidia. The schizont multiplies in number by binary fission (Figs. 10-14); the nuclear division is amitotic. In all the worms which we examined, we have not seen the condition commonly found in other Microsporidia where the schizogony is repeated rapidly and actively so



*Thelohania reniformis* nov. spec. Figs. 1-6, fresh spores. Figs. 7-8, stained spores (smears). Figs. 9-14, stages in schizogony (sections). Figs. 15-28, stages in sporogony (sections). Fig. 29, a spore mechanically pressed and stained after Fontana. Fig. 30, four infected epithelial cells of the host nematode (section). Figs. 1-29, × 2200; fig. 30, × 1560.

that the infected host cells usually become completely filled with the protozoan and even become deformed due to the enormous number of the parasites present in them. Schizogony in the present species is not very active and a slow multiplication by binary fission seems to be the only division process. There seem to exist some inhibiting factors counteracting the activity of the parasites.

The schizonts become the sporonts. Both the nucleus and the body become larger at the expense of the host cells. The cytoplasm of a sporont is more or less vacuolated. The nucleus divides three times, producing eight daughter nuclei and the cytoplasm of the sporont becomes divided into eight sporoplasts (Figs. 15-26). Each sporoblast develops into a spore (Figs. 27, 28). The membrane that surrounds the eight spores thus formed is distinctly seen even after the spores are fully matured.

Since previous authors have seen only stages which appear to be spores of organisms of doubtful nature from *Ascaris mystax,* we are unable to compare them with the forms we found. As the dimensions of the spores of the present form are however entirely different from any of the forms observed in the nematode of the cat and furthermore as the species under consideration has been found in the nematode of the mouse, we believe that we are justified in maintaining the species as hitherto unrecorded. The nucleus of the host cell infected by the parasite is similar in its location and structure to that of the uninfected host cell. The parasite does not seem to cause any pathological changes upon the host organ.

## SUMMARY

The records of the parasites of nematodes are reviewed.

A new microsporidian, *Thelohania reniformis,* parasitic in the epithelial cells of the intestine of a nematode, *Protospirura muris* parasitic in the common house mouse, *Mus musculus,* is described.

The schizonts multiply by a binary fission. The sporont develops into sporoblasts and finally into eight spores.

The nucleus of the host cell is not affected by the infection.

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